DIAGNOSTIC HYBRIDS, INC.
350 West State Street
Athens, OHIO 45701

DIAGNOSTIC HYBRIDS
Integrating Science and Humanity

K063675

SEP 24 2007

510(k) SUMMARY

D3 HERPES SIMPLEX VIRUS IDENTIFICATION KIT

Applicant

DIAGNOSTIC HYBRIDS, INC.
350 West State Street
Athens, OHIO 45701

Contact Information

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Date of preparation of 510(k) summary:

September 21, 2007 (revised)

Device Name

Trade name – Diagnostic Hybrids' D3 DFA Chlamydiae Culture Confirmation Kit
Common name – Fluorescent antibody test for Chlamydiae
Classification name Antiserum, fluorescent, Chlamydia trachomatis (21 CFR 866.3120, product code LJP)

Legally marketed devices to which equivalence is claimed:

K864389	American Microscan (distributed by Trinity Biotech) DFA Chlamydia Detection Kit
K864663	Kallestad (distributed by BioRad) Pathfinder® Chlamydia Culture Confirmation Kit/System
K895839	Diagnostic Products, Corp. (distributed by Remel) PathoDx® Chlamydia Culture Confirmation Kit

Device Description

The Diagnostic Hybrids' D3 DFA Chlamydiae Culture Confirmation Kit includes a Chlamydiae DFA Reagent that contains a blend of two murine MAbs directed against epitopes on the lipopolysaccharide of Chlamydiae. The kit is used for Chlamydiae detection in cell cultures of patient specimens.

Kit Components:

- Chlamydiae DFA Reagent - one dropper bottle containing fluorescein labeled murine MAbs directed against Chlamydiae. The buffered, stabilized, aqueous solution contains Evan's Blue as a counter-stain and 0.1% sodium azide as preservative.
- Chlamydiae Antigen Control Slides - 10 slides. Five individually packaged control slides with wells containing cell culture-derived Chlamydia trachomatis positive cells and five individually packaged control slides with wells containing cell culture-derived negative cells. Each slide is intended to be stained only one time.
- PBS Concentrate - a 40X concentrate consisting of 4% sodium azide (after dilution to 1X in water, the concentration of sodium azide in the solution is 0.1%) in PBS.
- Mounting Fluid - an aqueous, buffered, stabilized solution of glycerol and 0.1% sodium azide.

The cell cultures to be tested are fixed in acetone. The Chlamydiae DFA Reagent is added to the cells to determine the presence of chlamydial antigens (LPS). After incubating at 35°C to 37°C, the stained cells are rinsed with the diluted PBS Concentrate, a drop of the supplied Mounting Fluid is added and the monolayer is examined for presence of fluorescent inclusions using a fluorescence microscope equipped with the correct filter combination for FITC at a magnification of 100-400X.

Chlamydiae infected cells will be stained with bright apple-green fluorescence while uninfected cells will contain no apple-green fluorescence but will fluoresce red by the Evan's Blue counter-stain[1] which is included in the Chlamydiae DFA Reagent.

If no fluorescent cells are found, report result as, "No Chlamydiae detected". If fluorescent cells are found, indicating a Chlamydiae-positive specimen, report result as, "Chlamydiae isolated by cell culture."

Included in the kit are Chlamydiae Antigen Control Slides. A Control Slide is intended to function as an indicator that the kit reagents are working properly in the test. [The slides are prepared with either Chlamydiae infected cells (Positive Antigen Control Slide) or uninfected cells (Negative Antigen Control Slide).] Positive and negative controls must demonstrate appropriate staining characteristics for specimen results to be valid. Controls may also aid in the interpretation of test results.

It is recommended that cell culture positive (infected with known Chlamydiae isolate) and negative (uninfected cells) controls be run with each assay to provide a means to ensure adequate performance of the cell culture system used. If control cultures fail to perform correctly, results are considered invalid.

Intended Use

The Diagnostic Hybrids' D3 DFA Chlamydiae Culture Confirmation Kit is intended for the qualitative detection of Chlamydiae lipopolysaccharide (LPS) in inoculated cell cultures by immunofluorescence using fluoresceinated monoclonal antibodies (MAbs).

Performance has not been established with direct patient specimens.

Explanation

The test kit uses Chlamydiae antigen-specific murine MAbs that are directly labeled with fluorescein for rapid detection and identification of Chlamydiae.

The cell cultures to be tested are fixed in acetone. The Chlamydiae DFA Reagent is added to the cells to determine the presence of chlamydial antigens. After incubating at 35°C to 37°C, the stained cells are rinsed with the diluted PBS Concentrate, a drop of the supplied Mounting Fluid is added and a coverslip is placed on the prepared cells. The cells are examined using a fluorescence microscope.

Technological Characteristics

Fundamental technology and intended use of the device are similar as those of the predicate devices, which are based on a standard immunofluorescence assay technique using cells inoculated with patient specimens. They employ directly labeled fluorescein monoclonal antibodies specific for Chlamydiae antigens enabling visualization of the infected cells. A summary is provided in the table below:

Chlamydiae Test Systems	DFA	Direct Specimens	Culture Confirmation	FITC Label	Monoclonal Antibody	Target antigen (epitopes of the Chlamydiae lipopolysaccharide)
Diagnostic Hybrids	Yes	No	Yes	Yes	Yes	Yes
American Microscan	Yes	No	Yes	Yes	Yes	Yes
Kallestad	Yes	No	Yes	Yes	Yes	Yes
Diagnostic Products, Corp.	Yes	No	Yes	Yes	Yes	Yes

Non-clinical Performance

Staining patterns of the conjugated monoclonal antibodies on Chlamydiae infected cells were similar to those of the predicate devices.

The D3 DFA Chlamydiae Culture Confirmation Kit was characterized for its ability

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to specifically detect Chlamydiae species. The FITC-conjugated MAbs used in the kit exhibited characteristic staining patterns on Chlamydiae infected cells, with fluorescent inclusions in infected cells. The conjugated MAbs reacted with *Chlamydophila pneumoniae*, *Chlamydophila psittaci*, as well as 15 serovars of *Chlamydia trachomatis*.

The D^3 DFA Chlamydiae Culture Confirmation Kit was tested for cross-reactivity against a wide variety of cells and microorganisms. No cross-reactivity was observed for 20 host culture cell types or for 57 virus strains (cultured and processed for staining). Twenty-five (25) bacterial cultures, as well as one protozoan and one yeast specimen, were stained and examined for cross-reactivity. The protein-A produced by Staphylococcus aureus bound the Fc portion of the MAb's and appeared as small points of fluorescence while all other bacterial cultures were negative.

To test this product against Chlamydiae species, cell cultures (McCoy, BGMK, or HEp-2) were inoculated with approximately 1.5 x 106 infective units of *Chlamydia trachomatis*, *Chlamydophila psittaci*, or *Chlamydophila pneumoniae*, and incubated for 2 days to yield a 3+ to 4+ infection. Cultures were processed and stained with the Chlamydia DFA Reagent or specially prepared DFA reagents containing only one of the Chlamydia MAbs.

Stringent conditions for cross-reactivity testing were achieved by using a high concentration Chlamydiae DFA Reagent and high titers of microorganisms. The DFA was prepared at 1.5X the concentration that is provided in the kit.

Depending on the particular bacteria, the number of CFU tested ranged from $6.4x10^4$ to $2.9x10^7$. Depending on the virus, 150 to 2100 $TCID_{50}$ viruses were inoculated into shell vial culture and incubated for 24 to 48 hours, to yield a 1+ to 3+ infection, processed and stained with the 1.5X DFA according to the procedure detailed in the product insert. Additionally, for some viruses and other microorganisms, commercial slides containing the particular agent were used to test for cross reactivity.

Cell cultures were prepared in shell vial format. Confluent monolayers were stained with the 1.5X DFA Reagent according to the procedure as detailed in this product insert, then examined for cross reactivity.

Organisms and cell lines which were tested against the Chlamydiae DFA Reagent are listed below.

Results of Specificity and Cross Reactivity Testing

MICROORGANISMS (BACTERIA, YEAST, PROTOZOA)			
CHLAMYDIAE	**Serovar**	**Inoculum (Infective units per culture)**	**Result (Reactive = +) (Negative = -)**
Chlamydia trachomatis	A	1.5×10^6	+
	B	1.5×10^6	+
	C	1.5×10^6	+
	D	1.5×10^6	+
	E	1.5×10^6	+
	F	1.5×10^6	+
	G	1.5×10^6	+
	H	1.5×10^6	+
	I	1.5×10^6	+
	J	1.5×10^6	+
	K	1.5×10^6	+
	L1	1.5×10^6	+
	L2	1.5×10^6	+
	L3	1.5×10^6	+
	Ba	1.5×10^6	+
Chlamydophila pneumoniae		1.5×10^6	+
Chlamydophila psittaci		1.5×10^6	+

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OTHER BACTERIA		CFU Tested	Result (Reactive = +) (Negative = -)
Acholeplasma laidlawi		~1.0 x 10^7	-
Acinetobacter calcoaceticus		9.7 x 10^5	-
Bordetella bronchiseptica		1.8 x 10^5	-
Bordetella pertussis		4.7 x 10^6	-
Corynebacterium diphtheriae		2.5 x 10^6	-
Escherichia coli		2.6 x 10^5	-
Gardnerella vaginalis		5.0 x 10^5	-
Haemophilis influenzae type A		9.3 x 10^5	-
Klebsiella pneumoniae		6.4 x 10^6	-
Legionella pneumophila		6.5 x 10^4	-
Moraxella cartarrhalis		6.4 x 10^4	-
Mycoplasma hominis		~1.0 x 10^4	-
Mycoplasma orale		~1.0 x 10^4	-
Mycoplasma pneumoniae		~1.0 x 10^4	-
Mycoplasma salivarium		~1.0 x 10^7	-
Neisseria gonorrhoeae		1.3 x 10^6	-
Proteus mirabilis		2.1 x 10^6	-
Pseudomonas aeruginosa		1.0 x 10^3	-
Salmonella enteriditis		2.5 x 10^6	-
Salmonella typhimurium		1.8 x 10^6	-
Staphylococcus aureus		1.0 x 10^7	+
Streptococcus agalactiae		9.6 x 10^5	-
Streptococcus pneumoniae		8.0 x 10^5	-
Streptococcus pyogenes		2.9 x 10^7	-
Ureaplasma urealyticum		~1.0 x 10^4	-
YEAST		CFU Tested	Result (Reactive = +) (Negative = -)
Candida glabrata		8.7 x 10^6	-
PROTOZOAN		CFU Tested	Result (Reactive = +) (Negative = -)
Trichomonas vaginalis		Commercially available control slide[a]	-
VIRUS STRAINS		Inoculum ($TCID_{50}$)	Result (Reactive = +) (Negative = -)
Adenovirus	Type 1	725	-
	Type 3	725	-
	Type 6	725	-
	Type 7	725	-
	Type 8	725	-
	Type 10	725	-
	Type 13	725	-
	Type 14	725	-
	Type 18	725	-
	Type 31	725	-
	Type 40	725	-
	Type 41	725	-
Influenza A	Aichi	2.1 x 10^3	-
	Malaya	2.1 x 10^3	-

[a] Test material is from commercially available prepared slides. Each positive well contains 10 to 50% reactive cells.

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	Hong Kong	2.1×10^3	-
	Denver	2.1×10^3	-
	Port Chalmers	2.1×10^3	-
	PR	2.1×10^3	-
	Victoria	2.1×10^3	-
Influenza B	Hong Kong	2.1×10^3	-
	Maryland	2.1×10^3	-
	Mass	2.1×10^3	-
	Taiwan	2.1×10^3	-
	GL	2.1×10^3	-
	Russia	2.1×10^3	-
RSV	Long	2.1×10^3	-
	Wash	2.1×10^3	-
	9320	2.1×10^3	-
Parainfluenza 1	C-35	Commercially available control slide[a]	-
Parainfluenza 2	Greer	Commercially available control slide[a]	-
Parainfluenza 3	C 243	Commercially available control slide[a]	-
HSV-1	1F	150	-
	MacIntyre	150	-
HSV-2	MS	150	-
	Strain G	150	-
CMV	Towne	700	-
	AD169	700	-
	Davis	700	-
VZV	Ellen	500	
Echovirus	4, 6, 9, 11, 30, 34	Commercially available control slide[a]	-
Coxsackievirus	B1, B2, B3, B4, B5, B6	Commercially available control slide[a]	-
Mumps		Commercially available control slide[a]	-
Measles (Rubeola)		Commercially available control slide[a]	-
Poliovirus	Types 1, 2, 3	Commercially available control slide[a]	-
Epstein-Barr		Commercially available control slide[a]	-

Clinical Performance

Clinical studies included nine hundred and ninety four (994) original specimens evaluated for the presence of Chlamydiae by this product ("Subject" test) and three currently marketed Culture Confirmation Kits ("Comparison" tests). These evaluations were conducted at three external laboratory sites using one Comparison Device (American Microscan DFA Chlamydia Detection Kit) and one in-house laboratory where the two other Comparison Devices were used: (1) A reference laboratory in the southeastern United States; (2) A hospital laboratory in the mid-west United States; (3) A hospital laboratory in the southwestern United States; and (4) Diagnostic Hybrids' in-house virology laboratory.

A summary of the specimens by Site is presented in the table below.

Number of Specimens in Each Study Site

	Fresh prospective	Frozen prospective	Archived (frozen)	Site Total
1	156	240	0	396
2	90	84	26	200
3	23	68	187	278
4	0	120	0	120

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Percent Agreement and 95% Confidence Interval between the Subject Device and Comparison test for the three external laboratory sites was calculated and is presented in the table below (results from both fresh and frozen specimens are included).

Percent Agreement of All Tests

		Comparison Device	
		+	–
Subject Device Diagnostic Hybrids	+	42	4
	–	2	613
Positive Percent Agreement[b] (PPA)		95.5%	
95% CI[c]- PPA		84.5%-99.4%	
Negative Percent Agreement[d] (NPA)		99.4%	
95% CI – NPA		98.3%-99.8%	

[1] Specter, S., Hodinka, R. L., and Young, S.A. 2000, Clinical Virology Manual, Washington D.C., ASM Press, 420-424.

[b] "Positive Percent Agreement", or "PPA", values were calculated according to {[Total Number of Positive Results in Agreement by both Subject and Comparison Tests) divided by [(Total Number of Positive Results in Agreement by both Subject and Comparison Tests) plus (Number of Results Positive by Comparison but Negative by Subject)]} multiplied by 100%.

[c] "95% CI" refers to 95% Confidence Intervals, which were calculated according to Exact method (Clopper, C. and S. Pearson, Biometrika 26:404-413, 1934).

[d] "Negative Percent Agreement", or "NPA", values were calculated according to {[Total Number of Negative Results in Agreement by both Subject and Comparison Tests) divided by [(Total Number of Negative Results in Agreement by both Subject and Comparison Tests) plus (Number of Results Negative by Comparison but Positive by Subject)]} multiplied by 100%.

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DEPARTMENT OF HEALTH & HUMAN SERVICES

Public Health Service

Food and Drug Administration
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Rockville MD 20850

SEP 2 4 2007

Ms. Gail R. Goodrum
Vice President, Regulatory and Quality Affairs
Diagnostic Hybrids, Inc.
350 West State Street
Athens, OH 45701

Re: k063675
Trade/Device Name: Diagnostic Hybrids' D^3 Chlamydiae Culture Confirmation Kit
Regulation Number: 21 CFR § 866.3120
Regulation Name: Chlamydia serological reagents
Regulatory Class: I
Product Code: LJP
Dated: August 10, 2007
Received: August 13, 2007

Dear Ms. Goodrum:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration.

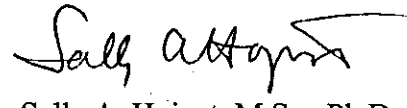
If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to such additional controls. Existing major regulations affecting your device can be found in Title 21, Code of Federal Regulations (CFR), Parts 800 to 895. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Parts 801 and 809); and good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820).

This letter will allow you to begin marketing your device as described in your Section 510(k) premarket notification. The FDA finding of substantial equivalence of your device to a legally marketed predicate device results in a classification for your device and thus, permits your device to proceed to the market.

If you desire specific advice for your device on our labeling regulation (21 CFR Part 801), please contact the Office of In Vitro Diagnostic Device Evaluation and Safety at 240-276-0450. Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21CFR Part 807.97). For questions regarding postmarket surveillance, please contact CDRH's Office of Surveillance and Biometric's (OSB's) Division of Postmarket Surveillance at 240-276-3474. For questions regarding the reporting of device adverse events (Medical Device Reporting (MDR)), please contact the Division of Surveillance Systems at 240-276-3464. You may obtain other general information on your responsibilities under the Act from the Division of Small Manufacturers, International and Consumer Assistance at its toll-free number (800) 638-2041 or (240) 276-3150 or at its Internet address http://www.fda.gov/cdrh/industry/support/index.html.

Sincerely yours,



Sally A. Hojvat, M.Sc., Ph.D.
Director
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Enclosure

Indications for Use

510(k) Number (if known): K063675

Device Name: Diagnostic Hybrids' D^3 DFA Chlamydiae Culture Confirmation Kit

Indications for Use: The Diagnostic Hybrids' D^3 DFA Chlamydiae Culture Confirmation Kit is intended for the qualitative detection of Chlamydiae lipopolysaccharide (LPS) in inoculated cell cultures by immunofluorescence using fluoresceinated monoclonal antibodies (MAbs).
Performance has not been established with direct patient specimens.

Prescription Use ___X___ (Part 21 CFR 801 Subpart D) AND/OR Over-The-Counter Use _____ (21 CFR 801 Subpart C)

(PLEASE DO NOT WRITE BELOW THIS LINE-CONTINUE ON ANOTHER PAGE OF NEEDED)

Concurrence of CDRH, Office of Device Evaluation (ODE)

Division Sign-Off

Office of In Vitro Diagnostic Device Evaluation and Safety

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